FINAL REPORT

Microbiological Sampling Report

for

National Oceanic & Atmospheric Administration

A Sampling Conducted on the Eleventh Floor
of Building SSMC-4
on June 21, 2000

Interagency Agreement #: D8H00CO31200
Task: 9903

January 31, 2001

Prepared by
US Public Health Service

Division of Federal Occupational Health

Bethesda Central Office

Executive Summary

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in a room 11125 of Building SSMC-4, located at 1305 East-West Highway, Silver Spring, Maryland. This sampling was conducted on June 21, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from this room and an indoor reference room 11132. Air samples were also collected from outdoors.

Findings are as follows:

· Indoor airborne fungal levels, by Andersen sampling, and indoor spore levels, by Zefon sampling, were lower than those of outdoors.

· *Stachybotrys chartarum* was not detected from any air, swab, or contact plate samples.

· In general, fungal burden on hard, horizontal surfaces of both rooms was low.

· Very low fungal burden was detected from swab samples collected from surfaces of supply diffusers and return troughers in light fixture.

· The fungal level in carpet dust of indoor reference room 11132 was higher than that of 11125 (10^4 vs. 10^3 CFU/g of fine dust). *Stachybotrys chartarum* was detected from carpet dust of room 11125.

· Fungal levels in furniture dust of both rooms were at 10^4 CFU/g of fine dust levels. *Stachybotrys chartarum* was not detected from any furniture dust sample.

INTRODUCTION

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in a room 11125 of Building SSMC-4, located at 1305 East-West Highway, Silver Spring, Maryland. This sampling was conducted on June 21, 2000. Air

(both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from this room and an indoor reference room 11132. Air samples were also collected from outdoors.

EVALUATION METHODOLOGY

Various types of samples were collected from these rooms on June 21, 2000.

Air Samples

Two types of air samples were collected from each room: (1) culturable method using Andersenâ N-6 samplers at a flow rate of 28.3 L/min, and (2) non-culturable method using Zefonâ Air-O-Cell cassettes at a flow rate of 15 L/min. Indoor Andersenâ air samples were collected for 3 minutes and outdoor samples were collected for both one and three minutes. Two percent (2 %) malt extract agar (MEA) and cellulose Czapek agar (CCA) was used to recover general fungi and cellulose-loving fungi, respectively. Non-culturable air samples were collected at the aforementioned sampling locations. Indoor samples were collected for ten minutes and outdoor samples were collected for both five and ten minutes. Outdoor air samples were collected near the entrance of the building. Temperature and relative humidity measurements were collected from room 11125 by a battery operated, direct readout Hygroskopâ meter.

Contact Plate Samples

To determine fungal burden on various surfaces of these rooms, four contact plate samples were collected from each room. Samples were collected from randomly selected hard, horizontal surfaces. Sampling was conducted by pressing the MEA-filled Rodacâ plate against the surface of interest for five seconds. A total of eight contact plate samples were collected.

Swab Samples

Swab samples were collected from surfaces of each supply diffusers and return troughers at the light fixture of each room. They were collected by wiping a known area of surface with a sterile cotton swab (Culturetteâ) wetted with holding media. Approximately 5 in^2 area was wiped for return trougher and 4

in^2 for supply diffusers. The swab was then placed directly into its holder. Each holder was labeled with an identifiable number. A total of six wipe samples were collected from these rooms.

Vacuum Dust Samples

Dust accumulated on carpeting and chairs and fabric system furniture were collected with a High Efficiency Particulate Air (HEPA) vacuum attached with a special "sock" device. For each carpet sample, a 3-ft by 3-ft area was vacuumed for at least five minutes. Total surface areas of 9 ft^2 were vacuumed from system furniture and chairs, and composite as one sample. One carpet sample and one composite furniture sample were collected from each room.

All samples collected were sent for next morning delivery to FOH's Environmental Microbiology Laboratory (EML) in Philadelphia, Pennsylvania for analysis.

Laboratory Procedures

Upon receipt, all Andersenâ air and contact plate samples were incubated in a 25˚C incubator. Each swab sample was suspended in sterile distilled water, diluted serially, and inoculated onto agar plates. Both MEA and CCA were used for retrieving fungi. At least three dilution series were used for each sample. Each vacuum dust sample was sieved through a 250-mm sieve. The fine dust (< 250-mm) retrieved was then weighed and followed the dilution plating for fungal analysis.

All plates were incubated in a 25˚C incubator. They were examined every other day for up to 10 days to ensure the full recovery of fungi. Fungal identification was based on colony morphology, spores and conidia formation. Total fungal colonies formed on each MEA plate and *Stachybotrys chartarum* on CCA plates were counted and recorded. Fungal levels in samples were presented as colony forming units (CFUs) per measuring unit. For example, CFU/m^3 for Andersenâ air samples, CFU/in^2 for swab samples, CFU/plate for contact plate samples, and CFU/g of fine dust for vacuum dust samples.

All Zefonâ cassette samples were analyzed by the Environmental Microbiology Laboratory in Escondido, California for direct microscopic examination. Fungal spores were identified and their airborne levels were presented as $spores/m^3$.

RESULTS AND DISCUSSION

Temperature and Relative Humidity

Indoor temperature and relative humidity were recorded in room 11125 only. The respective reading was 75.0°F, and 40.0%.

Microbiological Analyses Results

All laboratory analytical results from FOH's EML are presented in a laboratory report #NOAA-00-46R-A (Attachment A). Results from direct microscopic examination of Zefonâ cassette samples are presented in Attachment B.

Air Samples

Andersen Results

Outdoor airborne fungal levels (10^2 CFU/m^3) were higher than those of indoors (35 CFU/m^3) (Table 1). *Cladosporium* dominated outdoor fungal flora. Other fungi recovered from outdoors were *Alternaria* Basidiomycetes, *Epicoccum*, Ascomycetes, *Paecilomyces*, and *Penicillium*. Fungi detected indoors were similar to those of outdoors. *Stachybotrys chartarum* was not detected from these samples.

Zefon Results

Outdoor fungal spore levels were higher than those of indoors (Table 1). Total fungal level in reference room was higher than room 11125 (274 vs. 54 spores/m^3) (Table 1). Basidiospores, Ascospores, and spores of *Cladosporium* dominated outdoor fungal spore flora. Other fungal spores detected from outdoors were *Alternaria*, *Epicoccum, Penicillium/ Aspergillus* types, and Smuts, Periconia, and Myxomycetes. Fungal spores detected indoors were similar to those of outdoors. *Stachybotrys chartarum* was not detected from any sample collected.

Table 1. Airborne fungal levels at different rooms of the 11th floor in SSMC-4 on June 21, 2000.

Rooms **Parameters**	**11132** **Reference#**	**11125**	**Outdoors**

Airborne Fungal Levels (CFU/m^3)	35	35	436* 742
Total Fungal Spores (Spores/m^3)	274	54	11,194* 7,840

Indoor reference.

* Two samples were collected from outdoors.

Swab Samples

Most (five out of six) samples collected from surfaces of supply diffusers and return troughers in light fixtures were below the detection limits (BDL) (3 CFU/in^2 for supply diffuser and 2 CFU/in^2 for return trougher). The only sample showing fungal growth was collected from a return trougher surface in room 11132 with a fungal level of 66 CFU/in^2. Yeast dominated this sample.

Contact Plate Samples

In general, low fungal levels, ranged from BDL of 1 CFU/plate to 6 CFU/plate, were detected from these samples.

Vacuum Dust Samples

Carpet Dust

The fungal level in the fine dust collected from the carpet of indoor reference room 11132 was at 10^4 CFU/g of fine dust level (Table 2) with *Aspergillus sp.* as predominant fungal genus recovered. A lower (10^3 CFU/g of fine dust) fungal level was detected from room 11125 (Table 2). *Cladosporium* dominated and *Stachybotrys chartarum* was detected from this sample.

Furniture Dust

Fungal levels in the furniture dust of these rooms were at 10^4 CFU/g of fine dust level (Table 2). *Aspergillus sp.* dominated the samples collected from room 11125 while *Cladosporium* dominated the sample collected from reference room 11132. *Stachybotrys chartarum* was not detected from these samples (Table 2).

Table 2. Total fungal levels (CFU/g of fine dust) in fine dust collected from carpet and furniture of rooms 11125 and 11132 of SSMC-4, by vacuum dust sampling, collected on June 21, 2000.

Rooms	**11132** **Reference#**	**11125**
Carpet **(CFU/g of fine dust)**	32,000 (-*)	3,200 (+)
Furniture **(CFU/g of fine dust)**	30,303 (-)	32,000 (-)

Indoor reference.

* +: *Stachybotrys chartarum* was detected on MEA and/or CCA plates.

-: *Stachybotrys chartarum* was not detected on MEA and CCA plates.

CONCLUSIONS

· Indoor airborne fungal levels, by Andersen sampling, and indoor spore levels, by Zefon sampling, were lower than those of outdoors.

· *Stachybotrys chartarum* was not detected from any air, swab, and contact plate samples.

· In general, fungal burden on hard, horizontal surfaces of these rooms was low.

· Very low fungal burden was detected from wipe samples collected from surfaces of supply diffusers and return troughers in light fixture.

· The fungal level in carpet dust of indoor reference room 11132 was higher than that of 11125 (10^4 vs. 10^3 CFU/g of fine dust). *Stachybotrys chartarum* was detected from carpet dust of room 11125.

· Fungal levels in furniture dust of both rooms were at 10^4 CFU/g of fine dust levels. *Stachybotrys chartarum* was not detected from these furniture dust samples.

RECOMMENDATIONS

· Conduct thorough cleaning of both rooms by HEPA vacuuming and wet wiping.

· Implement an emergency water intrusion protocol for this building to adequately manage any unexpected water intrusion in order to prevent fungal proliferation.

ATTACHMENT A

Microbiological laboratory report #NOAA-00-46R-A for samples

Collected from eleventh floor of SSMC-4, on June 21, 2000.

ATTACHMENT B

Results from microscopic examination of Zefon air samples collected

from eleventh floor of SSMC-4, on June 21, 2000.

USPHS DFOH ENVIRONMENTAL MICROBIOLOGY LABORATORY, PHILADELPHIA, PA

LABORATORY REPORT #NOAA-00-46R-A

Client agency: National Oceanic and Atmospheric Administration, Silver Spring, MD

POIS#/task #: D8H00CO31200 / 9903

Sampling date: 6/21/00

Dates of inoculation: 6/21/00 (air and contact plates), 6/22/00 (wipes), and 6/23/00 (dust)

General location: SSMC-4, Silver Spring, MD

Specific location: 11th floor

Sampling techniques: Air (Andersen N-6 sampler), contact plate, wipe, and vacuum dust samplings

Medium used: Malt extract agar (MEA) and Cellulose Czapek agar (CCA) for fungi

Samples submitted by: J. Sobelman

Date characterization completed: 7/4/00

(A) Air samples on MEA and CCA plates

Sample ID	Sampling Location	Air Volume (L)	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
A1	Room 11125	84.9	1. *Alternaria* (1*) 2. *Cladosporium* (1) 3. *Penicillium* (1) CFU/m^3 = 35	No
A2	Room 11132, control	84.9	1. *Epicoccum* (1) 2. *Penicillium* (1) 3. Basidiomycetes (1) CFU/m^3 = 35	No

OA1	Outside	84.9	1. *Cladosporium* (23) 2. *Epicoccum* (4) 3. *Alternaria* (3) 4. *Paecilomyces* (1) 5. Ascomycetes (2) 6. Basidiomycetes (4) CFU/m^3 = 436	No
Sample ID	**Sampling Location**	**Air Volume (L)**	**Fungi on MEA @ 25°C**	**Presence of *Stachybotrys chartarum**** on CCA @ 25º C**
OA2	Outside	28.3	1. *Cladosporium* (11) 2. *Alternaria* (6) 3. *Epicoccum* (1) 4. *Penicillium* (1) 5. Basidiomycetes (2) CFU/m^3 = 742	No
SB	Shipping blank	NA#	No fungal growth	No

(B) Contact plate samples on MEA plates

Sample ID	**Sampling Location**	**Fungi detected on MEA @ 25°C**
CP1	Room 11125, desk	1. *Cladosporium* (1) 2. sterile fungi (1) **CFU/plate = 2**
CP2	Room 11125, top of system furniture	1. *Cladosporium* (1) 2. Ascomycetes (1) **CFU/plate = 2**
CP3	Room 11125, top shelf	1. *Ulocladium* (1) **CFU/plate = 1**

CP4	Room 11125, top of file cabinet	1. *Cladosporium* (2) 2. *Ulocladium* (1) 3. sterile fungi (3) **CFU/plate = 6**
CP5	Room 11132, top of desk	1. *Penicillium* (1) 2. sterile fungi (2) **CFU/plate = 3**

Sample ID	**Sampling Location**	**Fungi detected on MEA @ 25°C**
CP6	Room 11132, top of system furniture	1. *Cladosporium* (1) **CFU/plate = 1**
CP7	Room 11132, table of fax machine	1. *Penicillium* (1) 2. sterile fungi (1) **CFU/plate = 2**
CP8	Room 11132, table over file	1. *Alternaria* (1) 2. *Cladosporium* (1) 3. sterile fungi (1) **CFU/plate = 3**

(C) Wipe samples on MEA and CCA plates

Sample ID	**Sampling Location**	**Area (in^2)**	**Dilution factor**	**Fungi on MEA @ 25°C**	**Presence of *Stachybotrys chartarum**** on CCA @ 25º C**
LC	Lab control	NA	10X-MEA 10X-CCA	No fungal growth	No
W1	Room 11125, supply 1	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No

W2	Room 11125, return 1	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W3	Room 11125, supply 2	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No
W4	Room 11125, return 2	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W4B	Room 11132, supply, control	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No

Sample ID	**Sampling Location**	**Area (in^2)**	**Dilution factor**	**Fungi on MEA @ 25°C**	**Presence of *Stachybotrys chartarum**** on CCA @ 25º C**
W5	Room 11132, return, control	5	10X-MEA 10X-CCA	1. *Aspergillus sp.* (1) 2. *Penicillium* (1) 3. yeast (31) CFU/in^2 = 66	No

(D) Vacuum dust samples on MEA and CCA plates

Sample ID	**Sampling Location**	**Weight (g)**	**Dilution factor**	**Fungi on MEA @ 25°C**	**Presence of *Stachybotrys chartarum**** on CCA @ 25º C**
V01	Room 11125, carpet	0.100	40X-MEA 10X-CCA	1. *Cladosporium* (3) 2. *Alternaria* (2) 3. *Aspergillus sp.* (1) 4. *Aureobasidium* (1) 5. *Penicillium* (1) **CFU/g = 3,200**	Yes (1) CFU/g = 100

V02	Room 11125, furniture	0.100##	400X-MEA 10X-CCA	1. *Aspergillus sp.* (8) 2. *Alternaria* (4) 3. *Aureobasidium* (2) 4. *Cladosporium* (1) 5. *Epicoccum* (1) CFU/g = 3.2×10^4	No

Sample ID	**Sampling Location**	**Weight (g)**	**Dilution factor**	**Fungi on MEA @ 25°C**	**Presence of *Stachybotrys chartarum**** on CCA @ 25° C**
V03	Room 11132, carpet	0.100	400X-MEA 10X-CCA	1. *Aspergillus sp.* (5) 2. *Aureobasidium* (1) 3. *Epicoccum* (1) 4. *Paecilomyces* (1) CFU/g = 3.2×10^4	No
V04	Room 11132, furniture	0.066##	400X-MEA 10X-CCA	1. *Cladosporium* (3) 2. *Alternaria* (2) 3. *Epicoccum* (2) 4. *Aureobasidium* (1) 5. *Bipolaris* (1) 6. Ascomycetes (1) CFU/g = 3.0×10^4	No

* Colony counts. *** Toxigenic fungi. # Not applicable.

5ml of sterilized distilled water were added instead of 10ml.